

CLAIMS

1. A process of preparing membrane vesicles from a biological sample, characterised in that it comprises at least one anion exchange chromatography treatment of the sample.

2. Process according to claim 1, characterised in that it comprises at least one strong anion exchange chromatography treatment step.

3. Process according to any of claims 1 and 2, characterised in that it comprises at least one anion exchange and gel permeation chromatography steps.

4. Process according to any of the above claims, characterised in that the biological sample is a biological fluid, a culture supernatant, a cell lysate or a pre-purified solution.

5. A process of preparing membrane vesicles from a biological sample, characterised in that it comprises at least:

b) a treatment of the sample to prepare a sample enriched with membrane vesicles, and

c) an anion exchange chromatography and/or gel permeation chromatography treatment of the sample.

6. Process according to claim 5, characterised in that it comprises:

a) the culture of a population of membrane vesicle (e.g. exosome) producing cells under conditions enabling the release of vesicles,

b) a membrane vesicle enrichment step, and

0980319-072601

5

10

15

20

25

c) an anion exchange chromatography and/or gel permeation chromatography treatment of the sample.

7. Process according to claims 5 or 6, characterised in that the enrichment step comprises a clarification stage, optionally followed by a concentration stage.

8. Process according to any one of claims 5 to 7, characterised in that the enrichment step comprises an affinity chromatography step, preferably on a dye.

10 9. Process according to claim 7 or 8, characterised in that the enrichment step comprises a low speed centrifugation step and/or a filtration.

15 10. Process according to any one of claims 7 to 9, characterised in that the enrichment comprises at least an ultrafiltration step, particularly tangential.

11. A process of preparing membrane vesicles, characterised in that it comprises the following steps :

20 a) the culture of a population of membrane vesicle (e.g. exosome) producing cells under conditions enabling the release of vesicles,

b) the treatment of the culture supernatant with at least one ultrafiltration or affinity chromatography step, to produce a biological sample enriched with membrane vesicles (e.g. with exosomes), and

25 c) an anion exchange chromatography and/or gel permeation chromatography treatment step of the biological sample.

09060319-172200

12. Process according to claim 11, characterised in that it further comprises a filtration step d) of the treated preparation.

13. Process according to any of the above claims, characterised in that the membrane vesicles have a diameter between approximately 60 and 90 nm.

14. Process according to any of the above claims, characterised in that the membrane vesicles are vesicles produced by antigen presenting cells, particularly dendritic cells, B lymphocytes, macrophages or mastocytes.

15. Process according to claim 14, characterised in that the membrane vesicles are vesicles produced by dendritic cells, particularly of human origin.

16. Process according to any of claims 1 to 13, characterised in that the membrane vesicles are vesicles produced by tumoral cells, particularly of human origin.

17. A process of preparing membrane vesicles, characterised in that it comprises the following steps:

- a) obtaining a population of dendritic cells,
- b) culturing the dendritic cells under conditions enabling the production of membrane vesicles, and
- c) purifying the membrane vesicles using a process comprising at least an anion exchange chromatography treatment.

18. A process of preparing membrane vesicles, characterised in that it comprises the following steps:

- a) obtaining a population of dendritic cells,

T09220767E06860

*mb
A27*

*mb
A27*

b) culturing the dendritic cells under conditions enabling the production of membrane vesicles,

c) treating the culture supernatant to produce a biological sample enriched with membrane vesicles,
 5 particularly by an ultrafiltration or affinity chromatography step, and

d) purifying the membrane vesicles using a process comprising at least an anion exchange and/or gel permeation chromatography step.

10 19. Process according to claim 17 or 18, characterised in that the dendritic cells are obtained from a biological sample from a subject, e.g. bone marrow or peripheral blood.

15 20. Process according to claims 17 to 19, characterised in that the dendritic cells are immature.

21. Process according to any of claims 17 to 20, characterised in that the dendritic cells are sensitised to an antigen, prior to the membrane vesicle production.

20 22. Process according to any of claims 17 to 21, characterised in that, during step b), the dendritic cells are cultured under conditions stimulating membrane vesicle production.

25 23. Use of anion exchange chromatography for the preparation or purification of membrane vesicles.

24. Use of affinity chromatography for the preparation or purification of membrane vesicles.

25. Composition comprising membrane vesicles prepared using the process according to any of claims 1 to 22.

09290319 072604

mb
A27

mb
A28

mb
A29